

Amendments to the Claims:

Please amend claims 1, 8, 20, 22, 25, 40 and 46 as follows, and please cancel claims 2, 3, 23, 24 and 29-39 without prejudice to continued prosecution. The claims and their status are shown below.

1. (Currently Amended) A method for detecting the presence or absence of *Bordetella pertussis* in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises contacting said sample with a pair of IS481 primers to produce an IS481 amplification product if a *B. pertussis* IS481 nucleic acid molecule is present in said sample, wherein said pair of IS481 primers comprises a first IS481 primer and a second IS481 primer, wherein said first IS481 primer consists of the sequence 5'-CCA GTT CCT CAA GGA CGC-3' (SEQ ID NO:1) and said second IS481 primer consists of the sequence 5'-GAG TTC TGG TAG GTG TGA GCG TA-3' (SEQ ID NO:2), wherein said hybridizing step comprises contacting said sample with a pair of IS481 probes, wherein said pair of IS481 probes comprises a first IS481 probe and a second IS481 probe, wherein said first IS481 probe consists of the sequence 5'-CAC CGC TTT ACC CGA CCT TAC CGC CCA C- 3' (SEQ ID NO:3) and said second IS481 probe consists of the sequence 5'-GAC CAA TGG CAA GGC TCG AAC GCT TCA TC-3' (SEQ ID NO:11), wherein the members of said pair of IS481 probes hybridize within no more than five nucleotides of each other, wherein a first IS481 probe of said pair of IS481 probes is labeled with a donor fluorescent moiety and a second IS481 probe of said pair of IS481 probes is labeled with a corresponding acceptor fluorescent moiety; and

detecting the presence or absence of fluorescence resonance energy transfer (FRET) between said donor fluorescent moiety of said first IS481 probe and said corresponding acceptor fluorescent moiety of said second IS481 probe,

wherein the presence of FRET is indicative of the presence of *B. pertussis* in said biological sample, and wherein the absence of FRET is indicative of the absence of *B. pertussis* in said biological sample.

2-3. (Canceled)

4. (Original) The method of claim 1, wherein the members of said pair of IS481 probes hybridize within no more than two nucleotides of each other.

5. (Original) The method of claim 1, wherein the members of said pair of IS481 probes hybridize within no more than one nucleotide of each other.

6. (Original) The method of claim 1, wherein said donor fluorescent moiety is fluorescein.

7. (Original) The method of claim 1, wherein said acceptor fluorescent moiety is selected from the group consisting of LCTM-Red 640, LCTM-Red 705, Cy5, and Cy5.5.

8. (Currently Amended) The method of claim 1, wherein said detecting step comprises exciting said biological sample at a wavelength absorbed by said donor fluorescent moiety and visualizing and/or measuring the intensity of light ~~wavelength~~ emitted by said acceptor fluorescent moiety at said wavelength.

9. (Original) The method of claim 1, wherein said detecting comprises quantitating said FRET.

10. (Original) The method of claim 1, wherein said detecting step is performed after each cycling step.

11. (Original) The method of claim 1, wherein said detecting step is performed in real-time.

12. (Original) The method of claim 1, wherein the presence of said FRET within 40 cycling steps is indicative of the presence of a *B. pertussis* infection in said individual.

13. (Original) The method of claim 1, wherein the presence of said FRET within 30 cycling steps is indicative of the presence of a *B. pertussis* infection in said individual.

14. (Original) The method of claim 1, wherein the presence of said FRET within 25 cycling steps is indicative of the presence of a *B. pertussis* infection in said individual.

15. (Original) The method of claim 1, further comprising preventing amplification of a contaminant nucleic acid.

16. (Original) The method of claim 15, wherein said preventing comprises performing said amplifying step in the presence of uracil.

17. (Original) The method of claim 16, wherein said preventing further comprises treating said biological sample with uracil-DNA glycosylase prior to a first amplifying step.

18. (Original) The method of claim 1, wherein said biological sample is selected from the group consisting of nasopharyngeal swabs, nasopharyngeal aspirates, and throat swabs.

19. (Original) The method of claim 1, wherein said cycling step is performed on a control sample.

20. (Currently Amended) The method of claim 19, wherein said control sample comprises ~~said~~ a portion of said IS481 nucleic acid molecule.

21. (Original) The method of claim 1, wherein said cycling step uses a pair of control primers and a pair of control probes, wherein said control primers and said control probes are other than said IS481 primers and IS481 probes, respectively, wherein a control amplification product is produced if control template is present in said sample, wherein said control probes hybridize to said control amplification product.

22. (Currently Amended) A method for detecting the presence or absence of *Bordetella parapertussis* in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises contacting said sample with a pair of IS1001 primers to produce an IS1001 amplification product if a *B. parapertussis* IS1001 nucleic acid molecule is present in said sample, wherein said pair of IS1001 primers comprises a first IS1001 primer and a second IS1001 primer, wherein said first IS1001 primer consists of the sequence 5'-GGC GAT ATC AAC GGG TGA-3' (SEQ ID NO:5) and said second IS1001 primer consists of the sequence 5'-CAG GGC AAA CTC GTC CAT C-3' (SEQ ID NO:6), wherein said hybridizing step comprises contacting said biological sample with a pair of IS1001 probes, wherein said pair of IS1001 probes comprises a first IS1001 probe and a second IS1001 probe, wherein said first IS1001 probe consists of the sequence 5'-GGT TGG CAT ACC GTC AAG A-3' (SEQ ID NO:12) and said second IS1001 probe consists of the sequence 5'-GCT GGA CAA GGC TCG-3' (SEQ ID NO:13), wherein the members of said pair of IS1001 probes hybridize within no more than five nucleotides of each other, wherein a first IS1001 probe of said pair of IS1001 probes is labeled with a donor fluorescent moiety and a second IS1001 probe of said pair of IS1001 probes is labeled with a corresponding acceptor fluorescent moiety; and
detecting the presence or absence of FRET between said donor fluorescent moiety of said first IS1001 probe and said corresponding acceptor fluorescent moiety of said second IS1001 probe,

wherein the presence of FRET is indicative of the presence of *B. paraptussis* in said biological sample, and wherein the absence of FRET is indicative of the absence of *B. paraptussis* in said biological sample.

23-24. (Canceled)

25. (Currently Amended) A method for detecting the presence or absence of *Bordetella pertussis* and/or *B. paraptussis* in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises contacting said sample with a pair of IS481 primers and a pair of IS1001 primers to produce an IS481 amplification product if a *B. pertussis* IS481 nucleic acid molecule is present in said sample and an IS1001 amplification product if a *B. paraptussis* IS1001 nucleic acid molecule is present in said sample, wherein said pair of IS481 primers comprises a first IS481 primer and a second IS481 primer, wherein said first IS481 primer consists of the sequence 5'-CCA GTT CCT CAA GGA CGC-3' (SEQ ID NO:1) and said second IS481 primer consists of the sequence 5'-GAG TTC TGG TAG GTG TGA GCG TA-3' (SEQ ID NO:2), wherein said pair of IS1001 primers comprises a first IS1001 primer and a second IS1001 primer, wherein said first IS1001 primer consists of the sequence 5'-GGC GAT ATC AAC GGG TGA-3' (SEQ ID NO:5) and said second IS1001 primer consists of the sequence 5'-CAG GGC AAA CTC GTC CAT C-3' (SEQ ID NO:6), wherein said hybridizing step comprises contacting said sample with a pair of IS481 probes and a pair of IS1001 probes, wherein said pair of IS481 probes comprises a first IS481 probe and a second IS481 probe, wherein said first IS481 probe consists of the sequence 5'-CAC CGC TTT ACC CGA CCT TAC CGC CCA C- 3' (SEQ ID NO:3) and said second IS481 probe consists of the sequence 5'-GAC CAA TGG CAA GGC TCG AAC GCT TCA TC-3' (SEQ ID NO:11), wherein said pair of IS1001 probes comprises a first IS1001 probe and a second IS1001 probe, wherein said first IS1001 probe consists of the sequence 5'-GGT TGG CAT ACC GTC AAG A- 3' (SEQ ID NO:12) and said second IS1001 probe consists of the sequence 5'-GCT GGA CAA GGC TCG-3' (SEQ ID NO:13), wherein the members of said pair of IS481 probes hybridize within no more than five nucleotides of each other and wherein the members of said pair of IS1001 probes hybridize within no more than five nucleotides of each other, wherein a first IS481 probe of said pair of IS481 probes is labeled with a donor fluorescent moiety and wherein

a second IS481 probe of said pair of IS481 probes is labeled with a corresponding acceptor fluorescent moiety, wherein a first IS1001 probe of said pair of IS1001 probes is labeled with a donor fluorescent moiety and wherein a second IS1001 probe of said pair of IS1001 probes is labeled with a corresponding acceptor fluorescent moiety; and

detecting the presence or absence of FRET between said donor fluorescent moiety of said first IS481 probe and said corresponding acceptor fluorescent moiety of said second IS481 probe and/or between donor fluorescent moiety of said first IS1001 probe and said corresponding acceptor fluorescent moiety of said second IS1001 probe,

wherein the presence of FRET is indicative of the presence of *B. pertussis* and/or *B. paraptussis* in said biological sample, and wherein the absence of FRET is indicative of the absence of *B. pertussis* or *B. paraptussis* in said biological sample.

26. (Original) The method of claim 25, further comprising:

determining the melting temperature between one or both of said IS481 probes and said IS481 amplification product and between one or both of said IS1001 probes and said IS1001 amplification product, wherein said melting temperature(s) confirms said presence or absence of *B. pertussis* in said sample and said presence or absence of *B. paraptussis* in said sample.

27. (Original) The method of claim 26, wherein said melting temperature(s) distinguish between *B. pertussis* and *B. paraptussis* in said sample.

28. (Original) The method of claim 25, wherein said acceptor fluorescent moiety of said second IS481 probe and said acceptor fluorescent moiety of said second IS1001 probe are different.

29-39. (Withdrawn)

40. (Currently Amended) A method for detecting the presence or absence of *B. pertussis* in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises contacting said sample with a pair of IS481 primers to produce an IS481 amplification product if a *B. pertussis* IS481 nucleic acid molecule is present in said sample, wherein said pair of IS481 primers comprises a first IS481 primer and a second IS481 primer, wherein said first IS481 primer consists of the sequence 5'-CCA GTT CCT CAA GGA CGC-3' (SEQ ID NO:1) and said second IS481 primer consists of the sequence 5'-GAG TTC TGG TAG GTG TGA GCG TA-3' (SEQ ID NO:2),

wherein said hybridizing step comprises contacting said sample with an IS481 probe, wherein the IS481 probe is labeled with a donor fluorescent moiety and a corresponding acceptor fluorescent moiety, wherein said IS481 probe is selected from the group consisting of a first IS481 probe consisting of the sequence 5'-CAC CGC TTT ACC CGA CCT TAC CGC CCA C-3' (SEQ ID NO:3) and a second IS481 probe consisting of the sequence 5'-GAC CAA TGG CAA GGC TCG AAC GCT TCA TC-3' (SEQ ID NO:11); and

detecting the presence or absence of fluorescence resonance energy transfer (FRET) between said donor fluorescent moiety and said acceptor fluorescent moiety of said IS481 probe, wherein the presence or absence of FRET is indicative of the presence or absence of *B. pertussis* in said sample.

41. (Original) The method of claim 40, wherein said amplification employs a polymerase enzyme having 5' to 3' exonuclease activity.

42. (Original) The method of claim 41, wherein said donor and acceptor fluorescent moieties are within no more than 5 nucleotides of each other on said probe.

43. (Original) The method of claim 42, wherein said acceptor fluorescent moiety is a quencher.

44. (Original) The method of claim 40, wherein said IS481 probe comprises a nucleic acid sequence that permits secondary structure formation, wherein said secondary structure formation results in spatial proximity between said donor and said acceptor fluorescent moiety.

45. (Original) The method of claim 44, wherein said acceptor fluorescent moiety is a quencher.

46. (Currently Amended) A method for detecting the presence or absence of *B. pertussis* in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a dye-binding step, wherein said amplifying step comprises contacting said sample with a pair of IS481 primers to produce an IS481 amplification product if a *B. pertussis* IS481 nucleic acid molecule is present in said sample, wherein said pair of IS481 primers comprises a first IS481 primer and a second IS481 primer, wherein said first IS481 primer consists of the sequence 5'-CCA GTT CCT CAA GGA CGC-3' (SEQ ID NO:1) and said second IS481 primer consists of the sequence 5'-GAG TTC TGG TAG GTG TGA GCG TA-3' (SEQ ID NO:2), wherein said dye-binding step comprises contacting said IS481 amplification product with a

nucleic acid binding dye; and detecting the presence or absence of binding of said nucleic acid binding dye to said amplification product,

wherein the presence of binding is indicative of the presence of *B. pertussis* in said sample, and wherein the absence of binding is indicative of the absence of *B. pertussis* in said sample.

47. (Original) The method of claim 46, wherein said nucleic acid binding dye is selected from the group consisting of SYBRGreenI[®], SYBRGold[®], and ethidium bromide.

48. (Original) The method of claim 47, further comprising determining the melting temperature between said IS481 amplification product and said nucleic acid binding dye, wherein said melting temperature confirms said presence or absence of said *B. pertussis*.